



## REMARKS

Applicants have amended the specification to correct typographical errors.

Claims 10, 22, 35, 48, 60, 73, 86 and 98 are cancelled without prejudice. Applicants reserve the right to pursue the cancelled Claims in a related application. Dependency of Claims 11, 23, 24, 36, 37, 49, 61, 62, 74, 75, 87 and 99 are accordingly changed.

Claims 29, 43, 52, 67, 77, 90 and 102 are amended. Support for the amendments to Claims 29 and 67 may be found, e.g., in page 31 of the specification. Support for the amendment to Claims 52 may be found, e.g., in page 29 of the specification. The other claim amendments are typographical error corrections. Figure 6 is amended to correct typographical errors. These amendments do not present new matter.

### A. Claim Rejections under 35 U.S.C. § 112

Claims 1-102 are rejected under 35 U.S.C. § 112, first paragraph, for alleged lack of enablement. Applicants respectfully disagree with the Examiner.

Claim 1 is directed to a statistical test for the difference between perfect match and mismatch intensities. The result of the test is used to indicate whether a transcript is present. The Office Action, while acknowledging that two suitable test statistics are provided in the specification, alleges that the specification does not provide guidance on selecting other test statistics. Claim 1 recites "a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values," not any test statistic. The specification (e.g., page 24) teaches that the test statistic can be the simple difference between perfect match intensities and mismatch intensities or variations. One example of such variation (Ryder's discrimination score) is also provided. Applicants respectfully submit that it is well within the skill of an ordinary artisan to develop additional suitable statistics (variations that indicate the intensity difference) without

undue experimentation. In addition, the specification provides sufficient directions (statistics testing the intensity DIFFERENCE) and specific examples.

Another ground for the enablement rejection was the threshold value. Threshold values may depend on the specific requirements (e.g. stringency) of the test and applications. One of skill in the art would appreciate this variance in threshold values of a given test. It is well within the skill of an ordinary artisan to set threshold values for specific applications. In addition, the specification (e.g., pages 30 and 31), for example, provide ample guidance on associated threshold values (see particularly, lines 1-3 of page 31).

The Examiner also alleges that claims 1 and 26 and many of the dependent claims lack critical features including the requirement for significance levels as well as the conclusions to be drawn (present, absent) from any particular p-value. Applicants respectfully disagree. The claims are directed to a method (or a computer software product) that employs any suitable threshold value. A specific recitation of a significance value is not necessarily a critical feature of the claimed invention.

Claims 10, 12, 14-25, 29, 35-63 and 77-102 are allegedly rejected under 35 U.S.C. § 112, second paragraph, for being indefinite.

Claim 10 is allegedly confusing as the first and second significance levels overlap. The Examiner has objected to claims 22, 35, 48, 60, 73, 86, and 98 on the same grounds. Applicants respectfully disagree but for the purpose of expediting prosecution, have canceled Claims 10, 22, 35, 48, 60, 73, 86 and 98 without prejudice. Applicants will pursue the canceled claims in a related application.

Claim 12 allegedly lacks antecedent basis in claim 1 for significance levels. Applicants respectfully disagree. Antecedent basis may be found in the preceding dependent claims.

Claim 1 is allegedly directed to determining presence and not indicating marginal detection, with reference to claims 24, 37, 50, 62, 75, 88 and 100. Applicants respectfully traverse the objection. “Indicating marginal detection” in e.g., dependent claims 24, 37, 50, 62, 75, 88 and 100, describes a particular embodiment of “determining presence of a transcript” recited in claim 1.

Claim 14 recites a testing statistic  $median((PM_i - MM_i)/(PM_i + MM_i))$ . The Examiner alleges that this appears to be an erroneous formula, inconsistent with similar formulas on pages 5, 7, 10 and Figure 6. Claim 90 is allegedly objected to for the same reason. Applicants submit that the formula on pages 5, 7, 10 and Figure 6 contain typographical errors and have amended the specification to correct the same, making the formula consistent with that recited in Claims 14 and 90.

Claim 29 recites the subscript “1”. This is allegedly inconsistent with the specification which recites the subscript “3” in this context (page 31). Claim 67 is similarly objected to. Applicants have amended Claims 29 and 67 to recite the subscript “3”.

Claim 39 is allegedly confusing in reciting “a computer readable media for storing said computer program codes” as the language allegedly does not make clear if the computer readable media is actually storing the code. Applicants respectfully traverse the objection. The use of the word “for” indicates that the computer readable media is

actually storing the code. The computer program codes are a part of the computer software product.

Claim 43 is allegedly confusing in its dependency on Claim 42. Applicants have amended Claim 43 to make it dependent on claim 40.

Claim 52 recites a testing statistic  $median((PM_i-MM_i)/(PM_iMM_i))$ . The Examiner alleges that this appears to be erroneous although it is consistent with the formula on page 33. The Examiner notes that as written, the value would always be 1 and it would be inconsistent with Figure 6. Applicants have amended Claim 52 and page 33 to address the objection. These amendments do not present new matter as they would be obvious to one skilled in the art.

Claims 77 and 102 are allegedly confusing in reciting “said logical step comprising” as it is inconsistent with the plurality of logical steps required. Applicants have amended claims 77 and 102 to address this objection.

Claims 90 and 91 are allegedly confusing in their dependency upon claims 76-77, respectively, since Claims 76 and 77 are not directed to systems, but to software products. Applicants have amended Claim 90 to address this objection, but respectfully disagree with the Examiner’s assertion about Claim 91. Applicants submit that there is no error in the dependency of Claim 91 on Claim 77 since both are directed to systems.

Claim 102 is allegedly confusing in reciting “A processor” and “A memory” in the middle of the claim. The Examiner notes that “A” should be lowercase. Applicants have amended Claim 102 to address this objection.

Applicants submit that for the above reasons, the claim rejections under 35 U.S.C. § 112 should be withdrawn.

**B. Claim Rejections under 35 U.S.C. § 103**

Claims 1, 39 and 77 are rejected under 35 U.S.C. 103 (a) as allegedly being unpatentable over Lockhart et al. (1996) in view of either Hogg et al. or Hollander et al.

Lockhart et al. teach quantitative analysis of the signal intensity of PM and MM pairs from an array hybridization experiment while Hogg et al. and Hollander et al. discuss the Wilcoxon's signed rank test and its use for non-parametric data. The Office Action indicates that the motivation to combine is that one of skill in the art would have been aware that hybridization data from nucleic acid arrays was non parametric data. Applicants respectfully submit that the Office Action fails to point out the basis of the allegation that one of ordinary skill in the art, at the time of the invention, would have known that hybridization data is nonparametric. The Office Action also fails to indicate how the nonparametric nature of hybridization data would lead to the employment of Wilconxon's tests for analyzing intensity differences. Therefore, the rejections under 35 U.S.C. § 103 should be withdrawn.

## CONCLUSION

For these reasons, Applicants believe the application is now in condition for allowance and should be passed to issue. If the Examiner feels that a telephone conference would in any way expedite the prosecution of the application, please do not hesitate to call the undersigned at (408) 731-5000.

The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account 01-0431.

If the Examiner has any questions pertaining to this application, the Examiner is requested to contact the undersigned attorney.

Respectfully submitted,



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Dated: May 5, 2003

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**VERSION WITH MARKINGS TO SHOW CHANGES  
MADE TO THE APPLICATION**

***In the Specification***

Please amend the following paragraph on page 4, lines 10-18:

In one aspect of the invention, computer implemented methods are used for determining whether a transcript is present in a biological sample. The methods include the step of providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for the transcript, where each of the  $PM_i$  is paired with one of the  $MM_i$ ; calculating a  $p$ -value using one sided Wilcoxon's signed rank test, where the  $p$ -value is for a null hypothesis that  $\theta$ =a threshold value and an alternative hypothesis that said  $\theta >$  the threshold value, wherein said  $\theta$  is a test statistic for intensity difference between the perfect match intensity values and mismatch intensity values; and indicating whether the transcript is present based upon the  $p$ -value.

Please amend the following paragraph on page 5, lines 12-15:

In some particularly preferred embodiments, the testing statistic is  $median((PM_i - MM_i)/(PM_i + MM_i))$ . In these embodiments, the threshold value is a constant. Typically, the threshold value is around 0.001 to 0.05. Most preferably, the threshold value is around 0.015.

Please amend the following paragraph on page 6, lines 8-18:

The presence, marginal presence or absence (detected, marginally detected or undetected) of a transcript may be called based upon the  $p$ -value and significance levels.

Significance levels,  $\alpha_1$  and  $\alpha_2$  may be set such that:  $0 < \alpha_1 < \alpha_2 < 0.5$ . Note that for the one-sided test, if null hypothesis is true, the most likely observed  $p$ -value is 0.5, which is equivalent to 1 for the two-sided test. Let  $p$  be the  $p$ -value of one-sided signed rank test. In preferred embodiments, if  $p < \alpha_1$ , a “detected” call can be made (i.e., the expression of the target gene is detected in the sample). If  $\alpha_1 \leq p < \alpha_2$ , a marginally detected call may be made. If  $p \geq \alpha_2$ , “undetected call” may be made. The proper choice of significance levels and the thresholds can reduce false calls. In some preferred embodiments,  $0 < \alpha_1 < \alpha_2 < 0.06$ . In some particularly preferred embodiments,  $\alpha_1$  is around 0.04 and  $\alpha_2$  is around 0.06.

Please amend the following paragraph on page 7, lines 11-16:

In some particularly preferred embodiments of the computer software products of the invention, the testing statistic is  $median((PM_i - MM_i)/(PM_i[-] + MM_i))$  and threshold value is a constant. The computer program product may contain code for accepting user’s selection or input of the threshold value. A default value may be used as well. Typically, the threshold value is around 0.001 to 0.05. In a particularly preferred embodiment, the threshold value is around 0.015.

Please amend the following paragraph on page 7, lines 17-22:

The presen[t]ce, marginal presence or absence (detected, marginally detected or undetected) of a transcript may be called based upon the  $p$  –value and significance levels.



Significance levels,  $\alpha_1$  and  $\alpha_2$  may be set such that:  $0 < \alpha_1 < \alpha_2 < 0.5$ . In preferred embodiments, if  $p < \alpha_1$ , a “detected” call can be made (i.e., the expression of the target gene is detected in the sample). If  $\alpha_1 \leq p < \alpha_2$ , a marginally detected call may be made. If  $p \geq \alpha_2$ , “undetected call” may be made. The proper choice of significance levels and the

Please amend the following paragraph on page 8, lines 3-10:

The computer software product may include computer program code for indicating that the transcript is present, absent or marginally absent. The computer program code, when executed, may indicate the result by causing the display of the result on a display device such as a screen. Alternatively, the result may be outputted into a file. In addition, the result may be temporarily stored in a computer memory device so that other computer program modules may access this result. In some preferred embodiments, the computer software products may include code to accept a user's selection of various significance levels.

Please amend the following paragraph on page 9, lines 5-12:

The computer software product may include computer program code for indicating that the transcript is present, absent or marginally absent. The computer program code, when executed, may indicate the result by causing the display of the result on a display device such as a screen. Alternatively, the result may be outputted into a file. In addition, the result may be temporarily stored in a computer memory device so that other computer program modules may access this result. In some preferred

embodiments, the computer software products may include code to accept a user's selection of various significance levels.

Please amend the following paragraph on page 9, lines 13-17:

In addition, systems for determining whether a transcript is present in a biological sample are also provided. The systems include a processor; and a memory being coupled to the processor, the memory storing a plurality of machine instructions that cause the processor to perform a plurality of logical steps when implemented by the processor; the logical steps include the method steps of the invention.

Please amend the following paragraph on page 14, lines 1-22:

Methods for making and using molecular probe arrays, particularly nucleic acid probe arrays are also disclosed in, for example, U.S. Patent Numbers 5,143,854, 5,242,974, 5,252,743, 5,324,633, 5,384,261, 5,405,783, 5,409,810, 5,412,087, 5,424,186, 5,429,807, 5,445,934, 5,451,683, 5,482,867, 5,489,678, 5,491,074, 5,510,270, 5,527,681, 5,527,681, 5,541,061, 5,550,215, 5,554,501, 5,556,752, 5,556,961, 5,571,639, 5,583,211, 5,593,839, 5,599,695, 5,607,832, 5,624,711, 5,677,195, 5,744,101, 5,744,305, 5,753,788, 5,770,456, 5,770,722, 5,831,070, 5,856,101, 5,885,837, 5,889,165, 5,919,523, 5,922,591, 5,925,517, 5,658,734, 6,022,963, 6,150,147, 6,147,205, 6,153,743, 6,140,044 and D430024, all of which are incorporated by reference in their entireties for all purposes. Typically, a nucleic acid sample is [a] labeled with a signal moiety, such as a fluorescent label. The sample is hybridized with the array under appropriate conditions. The arrays are washed or otherwise processed to remove non-hybridized sample nucleic acids. The

hybridization is then evaluated by detecting the distribution of the label on the chip. The distribution of label may be detected by scanning the arrays to determine fluorescence intensities distribution. Typically, the hybridization of each probe is reflected by several pixel intensities. The raw intensity data may be stored in a gray scale pixel intensity file. The GATC™ Consortium has specified several file formats for storing array intensity data. The final software specification is available at [[www.gatcconsortium.org](http://www.gatcconsortium.org)] the Consortium's website and is incorporated herein by reference in its entirety. The pixel intensity files are usually large. For example, a GATC™ compatible image file may be approximately 50 Mb if there are about 5000 pixels on each of the horizontal and vertical axes and if a two byte integer is used for every pixel intensity. The pixels may be grouped into cells (see, GATC™

Please amend the following paragraph on page 17, lines 9-15:

The embodiments of the invention will be described using GeneChip® high oligonucleotide density probe arrays (available from Affymetrix, Inc., Santa Clara, CA, USA) as exemplary embodiments. One of skill in the art would appreciate that the embodiments of the invention are not limited to high density oligonucleotide probe arrays. In contrast, the embodiments of the invention are useful for analyzing any parallel large scale biological analysis, such as those using nucleic acid probe arrays, protein arrays, etc.

Please amend the following paragraph on page 18, lines 1-7:

in several patents previously incorporated by reference. In such embodiments, a single square-shaped feature on an array contains one type of probe. Probes are selected to be specific against desired target. Methods for selecting probe sequences are disclosed in, for example, U.S. Patent Application Nos. 09/718,295, [Attorney Docket Number 3359, filed November 21, 2000;] 09/721,042, [Attorney Docket Number 3367, filed November 21, 2000,] and 60/252,617, [Attorney Docket Number 3373, filed November 21, 2000,] all incorporated herein by reference in their entireties for all purposes.

Please amend the following paragraph on page 20, lines 14-21:

Computer software products may be written in any of various suitable programming languages, such as C, C++, C# (Microsoft®), Fortran, Perl, MatLab (MathWorks[, [www.mathworks.com](http://www.mathworks.com)]), SAS, SPSS and Java. The computer software product may be an independent application with data input and data display modules. Alternatively, the computer software products may be classes that may be instantiated as distributed objects. The computer software products may also be component software such as Java Beans (Sun Microsystems), Enterprise Java Beans (EJB, Sun Microsystems), Microsoft® COM/DCOM (Microsoft®), etc.

Please amend the following paragraph on page 23, lines 5-11:

In some embodiments, Wilcoxon's signed rank test is used to analyze paired PM and MM probes. In a block of  $n$  probe pairs (also known as atoms, Figure 3) for detecting a gene (typically 10, 15, or 20 probe pairs). Each probe pair typically consists

of two cells, one has the sequence designed to be perfectly matching the target sequence and the other has the sequence designed to be mismatching the target sequence, preferably at only a single nucleotide location (usually at the center of the sequence segment).

Please amend the following paragraph on page 23, lines 12-20:

Let the  $i$ -th perfectly matching cell intensity be  $PM_i$  and the  $i$ -th mismatching cell intensity be  $MM_i$  ( $i=1, \dots, n$ ). All these data are positive numbers. As described above, in some embodiments, the hybridization of each probe may be reflected by several pixel intensities. In such embodiments, the cell intensity is derived from the pixel intensities. In preferred embodiments, around 60, 70, 75, 80, 85, or 90 percentile of intensities of inner pixels in a cell is used to represent the cell intensity. In a particularly preferred embodiment, the 75 percentile of intensities of inner pixels in a cell is used to represent the cell intensity and is saved in a CEL file together with the number of pixels and the standard deviation of intensities at these pixels.

Please amend the following paragraph on page 29, lines 3-12:

In some particularly preferred embodiments, the following three statistics of cell intensities can be used to make calls based on one sided Wilcoxon's signed rank test.

The null hypothesis is denoted  $H_0$  and alternative hypothesis  $H_1$ .

(1)  $H_0: \text{median}(PM_i - MM_i) = \tau_l$ ;

$$H_1: \text{median}(PM_i - MM_i) > \tau_l$$

$$(2) \quad H_0: \text{median } (PM_i - MM_i) / (PM_i + MM_i) = \tau_2;$$

$$H_1: \text{median } (PM_i - MM_i) / (PM_i \pm MM_i) > \tau_2;$$

$$(3) \quad H_0: \text{median } (PM_i - B_i) = \tau_3;$$

$$H_1: \text{median } (PM_i - B_i) > \tau_3;$$

Please amend the paragraph on page 32, lines 15-22:

The presence, marginal presence or absence (detected, marginally detected or undetected) of a transcript may be called based upon the  $p$ -value and significance levels (54-58). Significance levels,  $\alpha_1$  and  $\alpha_2$  may be set such that:  $0 < \alpha_1 < \alpha_2 < 0.5$ . Note that for the one-sided test, if null hypothesis is true, the most likely observed  $p$ -value is 0.5, which is equivalent to 1 for the two-sided test. Let  $p$  be the  $p$ -value of one sided signed rank test. In preferred embodiments, if  $p < \alpha_1$ , a “detected” call can be made (i.e., the expression of the target gene is detected in the sample). If  $\alpha_1 \leq p < \alpha_2$ , a marginally detected call may be made. If  $p \geq \alpha_2$ , “undetected call” may be made. The proper choice of significance

***In the Claims***

Please amend Claims 11, 23, 24, 29, 36, 37, 43, 49, 52, 61, 62, 67, 74, 75, 77, 87, 90, 99, 100 and 102 as follows:

11. (amended) The method of Claim [10]9 wherein said second significance level is 0.06.

23. (amended) The method of Claim [22]21 wherein said second significance level is 0.06.

24. (amended) The method of Claim [22]21 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

29. (amended) The method of Claim 27 wherein said threshold value is calculated using:  $\tau_{[1]3} = c_{[1]3} \sqrt{\text{median}(PM_i)}$  wherein said  $c_{[1]3}$  is a constant.

36. (amended) The method of Claim [35]34 wherein said second significance level is 0.06.

37. (amended) The method of Claim [35]34 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

43. (amended) The computer software product of Claim [42]40 wherein threshold value is calculated using:  $\tau_1 = c_1 \sqrt{\text{mean}(PM_i)}$  wherein said  $c_1$  is a constant.

49. (amended) The computer software product of Claim [48]47 wherein said second significance level is 0.06.

52. (amended) The computer software product of Claim 40 wherein said testing statistic is  $\text{median}((PM_i - MM_i)/(PM_{i[-]} \pm MM_i))$ .

61. (amended) The computer software product of Claim [60]59 wherein said second significance level is 0.06.

62. (amended) The computer software product of Claim [60]59 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said computer program code for indicating further comprises computer code for indicating that said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .



67. (amended) The computer software product of Claim 66 wherein said threshold value is calculated using:  $\tau_{[1]3} = c_{[1]3} \sqrt{\text{median}(PM_i)}$  wherein said  $c_{[1]3}$  is a constant.

74. (amended) The computer software product of Claim [73]72 wherein said second significance level is 0.06.

75. (amended) The computer software product of Claim [73]72 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said code for indicating further comprises code for indicating that said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

77. (amended) A system for determining whether a transcript is present in a biological sample

comprising:

a processor; and

a memory being coupled to the processor, the memory storing a plurality of machine instructions that cause the processor to perform a plurality of logical steps when implemented by the processor, said logical steps comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for the transcript, wherein each of the  $PM_i$  is paired with one of the  $MM_i$ ;

calculating a  $p$ -value using one-sided Wilcoxon's signed rank test,

wherein the  $p$ -value is for a null hypothesis that  $\theta$ =a threshold value and an

alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values; and  
indicating whether said transcript is present based upon said  $p$ -value.

87. (amended) The system of Claim [86]85 wherein said second significance level is 0.06.

90. (amended) The [system] computer software product of Claim 76 wherein said testing statistic is  $median((PM_i - MM_i)/(PM_i + MM_i))$ .

99. (amended) The system of Claim [98]97 wherein said second significance level is 0.06.

101. (amended) The system of Claim [98]97 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

102. (amended) A system for determining whether a transcript is present in a biological sample comprising:

[A]a processor; and

[A]a memory being coupled to the processor, the memory storing a plurality of machine instructions that cause the processor to perform a

plurality of logical steps when implemented by the processor; said logical steps comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and background intensity values ( $B_i$ ) for said transcript, wherein each of said  $PM_i$  is paired with one of said  $B_i$ ;

103. (new) A method for determining whether a transcript is present in a biological sample comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for at least 5000 transcripts, wherein the  $PM_i$  for each of said 5000 transcripts is paired with one of the  $MM_i$ ;

calculating a  $p$ -value using one-sided Wilcoxon's signed rank test, wherein the  $p$ -value is for a null hypothesis that  $\theta$ =a threshold value and an alternative hypothesis that said  $\theta$ > said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values; and

indicating whether said transcript is present based upon said  $p$ -value.

## CURRENT VERSION OF CLAIMS PENDING IN THE FILE

What is claimed is:

1. (original) A method for determining whether a transcript is present in a biological sample

comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for the transcript, wherein each of the  $PM_i$  is paired with one of the  $MM_i$ ;

calculating a  $p$ -value using one-sided Wilcoxon's signed rank test, wherein the  $p$ -value is for a null hypothesis that  $\theta$ =a threshold value and an alternative hypothesis that said  $\theta$ > said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values; and

indicating whether said transcript is present based upon said  $p$ -value.

2. (original) The method of Claim 1 wherein said testing statistic is  $median(PM_i - MM_i)$ .
3. (original) The method of Claim 2 wherein said threshold value is zero.
4. (original) The method of Claim 2 wherein said threshold value is calculated using:  $\tau_1 = c_1 \sqrt{median(PM_i)}$  wherein said  $c_1$  is a constant.

5. (original) The method of Claim 2 wherein threshold value is calculated  
using:  $\tau_1 = c_1 \sqrt{\text{mean}(PM_i)}$  wherein said  $c_1$  is a constant.
6. (original) The method of Claim 2 wherein said step of indicating comprises indicating  
said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).
7. (original) The method of Claim 6 wherein said significance level is 0.01-0.08.
8. (original) The method of Claim 7 wherein said first significance level is 0.04.
9. (original) The method of Claim 7 wherein said step of indicating further comprises  
indicating said transcript is absent if said  $p$  is greater than or equal to a second  
significance level ( $\alpha_2$ ).
11. (currently amended) The method of Claim 9 wherein said second significance level is  
0.06.
12. (original) The method of Claim 11 wherein said first significance level ( $\alpha_1$ ) is smaller  
than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript  
is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

13. (original) The method of Claim 12 where first significance level is 0.04 and second significance level is 0.06.

14. (original) The method of Claim 1 wherein said testing statistic is  $\text{median}((PM_i - MM_i)/(PM_i + MM_i))$ .

15. (original) The method of Claim 14 wherein said threshold value is a constant.

16. (original) The method of Claim 15 wherein said threshold value is around 0.001 to 0.05.

17. (original) The method of Claim 16 wherein said threshold value is around 0.015.

18. (original) The method of Claim 17 wherein said step of indicating comprises indicating said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

19. (original) The method of Claim 18 wherein said significance level is 0.01-0.08.

20. (original) The method of Claim 19 wherein said first significance level is 0.04.

21. (original) The method of Claim 20 wherein said step of indicating further comprises indicating said transcript is absent if said  $p$  is greater than a second significance level ( $\alpha_2$ ).

23. (currently amended) The method of Claim 21 wherein said second significance level is 0.06.

24. (currently amended) The method of Claim 21 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

25. (original) The method of Claim 24 where first significance level is 0.04 and second significance level is 0.06.

26. (original) A method for determining whether a transcript is present in a biological sample comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and background intensity values ( $B_i$ ) for said transcript, wherein each of said  $PM_i$  is paired with one of said  $B_i$ ;

calculating a  $p$  value using one sided Wilcoxon's signed rank test, wherein said  $p$  value is for a null hypothesis that  $\theta =$  a threshold value and an alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and background intensity values; and

indicating whether said transcript is present based upon said  $p$  value.

27. (original) The method of Claim 26 wherein said testing statistic is  $median(PM_i - B_i)$ .

28. (original) The method of Claim 27 wherein said threshold value is zero.

29. (currently amended) The method of Claim 27 wherein said threshold value is

calculated using:  $\tau_3 = c_3 \sqrt{median(PM_i)}$  wherein said  $c_3$  is a constant.

30. (original) The method of Claim 27 wherein threshold value is calculated

using:  $\tau_3 = c_3 \sqrt{mean(PM_i)}$  wherein said  $c_3$  is a constant.

31. (original) The method of Claim 27 wherein said step of indicating comprises

indicating said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

32. (original) The method of Claim 31 wherein said significance level is 0.01-0.08.

33. (original) The method of Claim 32 wherein said first significance level is 0.04.

34. (original) The method of Claim 31 wherein said step of indicating further comprises

indicating said transcript is absent if said  $p$  is greater than a second significance level

( $\alpha_2$ ).



36. (currently amended) The method of Claim 34 wherein said second significance level is 0.06.

37. (currently amended) The method of Claim 34 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

38. (original) The method of Claim 37 where first significance level is 0.04 and second significance level is 0.06.

39. (original) A computer software product comprising:

computer program code for inputting a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for a transcript, wherein each of said  $PM_i$  is paired with one of said  $MM_i$ ;

computer program code for calculating a  $p$  value using one sided Wilcoxon's signed rank test, wherein said  $p$  value is for a null hypothesis that  $\theta =$  a threshold value and an alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values;

computer program code for indicating whether said transcript is present based upon said  $p$  value; and

a computer readable media for storing said computer program codes.

40. (original) The computer software product of Claim 39 wherein said testing statistic is  $median(PM_i - MM_i)$ .

41. (original) The computer software product of Claim 40 wherein said threshold value is zero.

42. (original) The computer software product of Claim 40 wherein said threshold value is calculated using:  $\tau_1 = c_1 \sqrt{median(PM_i)}$  wherein said  $c_1$  is a constant.

43. (currently amended) The computer software product of Claim 40 wherein threshold value is calculated using:  $\tau_1 = c_1 \sqrt{mean(PM_i)}$  wherein said  $c_1$  is a constant.

44. (original) The computer software product of Claim 40 wherein said computer program code of indicating comprises computer program code for indicating that said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

45. (original) The computer software product of Claim 44 wherein said significance level is 0.01-0.08.

46. (original) The computer software product of Claim 45 wherein said first significance level is 0.04.

47. (original) The computer software product of Claim 46 wherein said computer code for indicating further comprises computer program code for indicating that said transcript is absent if said  $p$  is greater than or equal to a second significance level ( $\alpha_2$ ).

49. (currently amended) The computer software product of Claim 47 wherein said second significance level is 0.06.

50. (original) The computer software product of Claim 49 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said computer program code of indicating further comprises computer program code for indicating that said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

51. (original) The computer software product of Claim 50 where first significance level is 0.04 and second significance level is 0.06.

52. (currently amended) The computer software product of Claim 40 wherein said testing statistic is  $median((PM_i - MM_i)/(PM_i + MM_i))$ .

53. (original) The computer software product of Claim 52 wherein said threshold value is a constant.

54. (original) The computer software product of Claim 53 wherein said threshold value is around 0.001 to 0.05.

55. (original) The computer software product of Claim 54 wherein said threshold value is around 0.015.

56. (original) The computer software product of Claim 53 wherein said computer program code for indicating comprises computer program code for indicating that said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

57. (original) The computer software product of Claim 56 wherein said significance level is 0.01-0.08.

58. (original) The computer software product of Claim 57 wherein said first significance level is 0.04.

59. (original) The computer software product of Claim 57 wherein said computer program code for indicating further comprises computer program code for indicating said transcript is absent if said  $p$  is greater than or equal to a second significance level ( $\alpha_2$ ).

61. (currently amended) The computer software product of Claim 59 wherein said second significance level is 0.06.

62. (currently amended) The computer software product of Claim 59 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said computer program code for

indicating further comprises computer code for indicating that said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

63. (original) The computer software product of Claim 62 where first significance level is 0.04 and second significance level is 0.06.

64. (original) A computer software product comprising:

computer program code for providing a plurality of perfect match intensity values ( $PM_i$ ) and background intensity values ( $B_i$ ) for a transcript, wherein each of said  $PM_i$  is paired with one of said  $B_i$ ;

computer program code for calculating a  $p$  value using one sided Wilcoxon's signed rank test, wherein said  $p$ -value is for a null hypothesis that  $\theta =$  a threshold value and an alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and background intensity values; and

computer program code for indicating whether said transcript is present based upon said  $p$ -value; and

a computer readable media for storing said codes.

65. (original) The computer software product of Claim 64 wherein said testing statistic is  $median(PM_i - B_i)$ .

66. (original) The computer software product of Claim 65 wherein said threshold value is zero.

67. (currently amended) The computer software product of Claim 66 wherein said threshold value is calculated using:  $\tau_3 = c_3 \sqrt{\text{median}(PM_i)}$  wherein said  $c_3$  is a constant.

68. (original) The computer software product of Claim 66 wherein threshold value is calculated using:  $\tau_3 = c_3 \sqrt{\text{mean}(PM_i)}$  wherein said  $c_3$  is a constant.

69. (original) The computer software product of Claim 66 wherein said step of indicating comprises indicating said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

70. (original) The computer software product of Claim 69 wherein said significance level is 0.01-0.08.

71. (original) The computer software product of Claim 70 wherein said first significance level is 0.04.

72. (original) The computer software product of Claim 71 wherein said computer software code of indicating further comprises computer software code for indicating that

said transcript is absent if said  $p$  is greater than or equal to a second significance level ( $\alpha_2$ ).

74. (currently amended) The computer software product of Claim 72 wherein said second significance level is 0.06.

75. (currently amended) The computer software product of Claim 72 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said code for indicating further comprises code for indicating that said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

76. (original) The computer software product of Claim 75 where first significance level is 0.04 and second significance level is 0.06.

77. (currently amended) A system for determining whether a transcript is present in a biological sample comprising:

a processor; and

a memory being coupled to the processor, the memory storing a plurality of machine instructions that cause the processor to perform a plurality of logical steps when implemented by the processor, said logical steps comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for the transcript, wherein each of the  $PM_i$  is paired with one of the  $MM_i$ ;

calculating a  $p$ -value using one-sided Wilcoxon's signed rank test,  
wherein the  $p$ -value is for a null hypothesis that  $\theta =$  a threshold value and an  
alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test  
statistic for intensity difference between said perfect match intensity values and  
mismatch intensity values; and  
indicating whether said transcript is present based upon said  $p$ -value.

78. (original) The system of Claim 77 wherein said testing statistic is  $median(PM_i - MM_i)$

79. (original) The system of Claim 78 wherein said threshold value is zero.

80. (original) The system of Claim 78 wherein said threshold value is calculated  
using:  $\tau_i = c_i \sqrt{median(PM_i)}$  wherein said  $c_i$  is a constant.

81. (original) The system of Claim 78 wherein threshold value is calculated  
using:  $\tau_i = c_i \sqrt{mean(PM_i)}$  wherein said  $c_i$  is a constant.

82. (original) The system of Claim 78 wherein said step of indicating comprises  
indicating said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

83. (original) The system of Claim 82 wherein said significance level is 0.01-0.08.



84. (original) The system of Claim 83 wherein said first significance level is 0.04.

85. (original) The system of Claim 83 wherein said step of indicating further comprises indicating said transcript is absent if said  $p$  is greater than or equal to a second significance level ( $\alpha_2$ ).

87. (currently amended) The system of Claim 85 wherein said second significance level is 0.06.

88. (original) The system of Claim 87 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

89. (original) The system of Claim 88 where first significance level is 0.04 and second significance level is 0.06.

90. (currently amended) The computer software product of Claim 76 wherein said testing statistic is  $median((PM_i - MM_i)/(PM + MM_i))$ .

91. (original) The system of Claim 77 wherein said threshold value is a constant.

92. (original) The system of Claim 91 wherein said threshold value is around 0.001 to 0.05.

93. (original) The system of Claim 92 wherein said threshold value is around 0.015.

94. (original) The system of Claim 91 wherein said step of indicating comprises indicating said transcript is present if said  $p$  is smaller than a first significance level ( $\alpha_1$ ).

95. (original) The system of Claim 94 wherein said significance level is 0.01-0.08.

96. (original) The system of Claim 95 wherein said first significance level is 0.04.

97. (original) The system of Claim 96 wherein said step of indicating further comprises indicating said transcript is absent if said  $p$  is greater than a second significance level ( $\alpha_2$ ).

99. (currently amended) The system of Claim 97 wherein said second significance level is 0.06.

100. (currently amended) The system of Claim 97 wherein said first significance level ( $\alpha_1$ ) is smaller than said ( $\alpha_2$ ) and said step of indicating further comprises indicating said transcript is marginally detected if  $\alpha_1 \leq p < \alpha_2$ .

101. (original) The system of Claim 100 where first significance level is 0.04 and second significance level is 0.06.

102. (currently amended) A system for determining whether a transcript is present in a biological sample comprising:

a processor; and

a memory being coupled to the processor, the memory storing a plurality of machine instructions that cause the processor to perform a plurality of logical steps when implemented by the processor; said logical steps comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and background intensity values ( $B_i$ ) for said transcript, wherein each of said  $PM_i$  is paired with one of said  $B_i$ ;

calculating a  $p$  value using one sided Wilcoxon's signed rank test, wherein said  $p$  value is for a null hypothesis that  $\theta$ =a threshold value and an alternative hypothesis that said  $\theta >$  said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and background intensity values; and

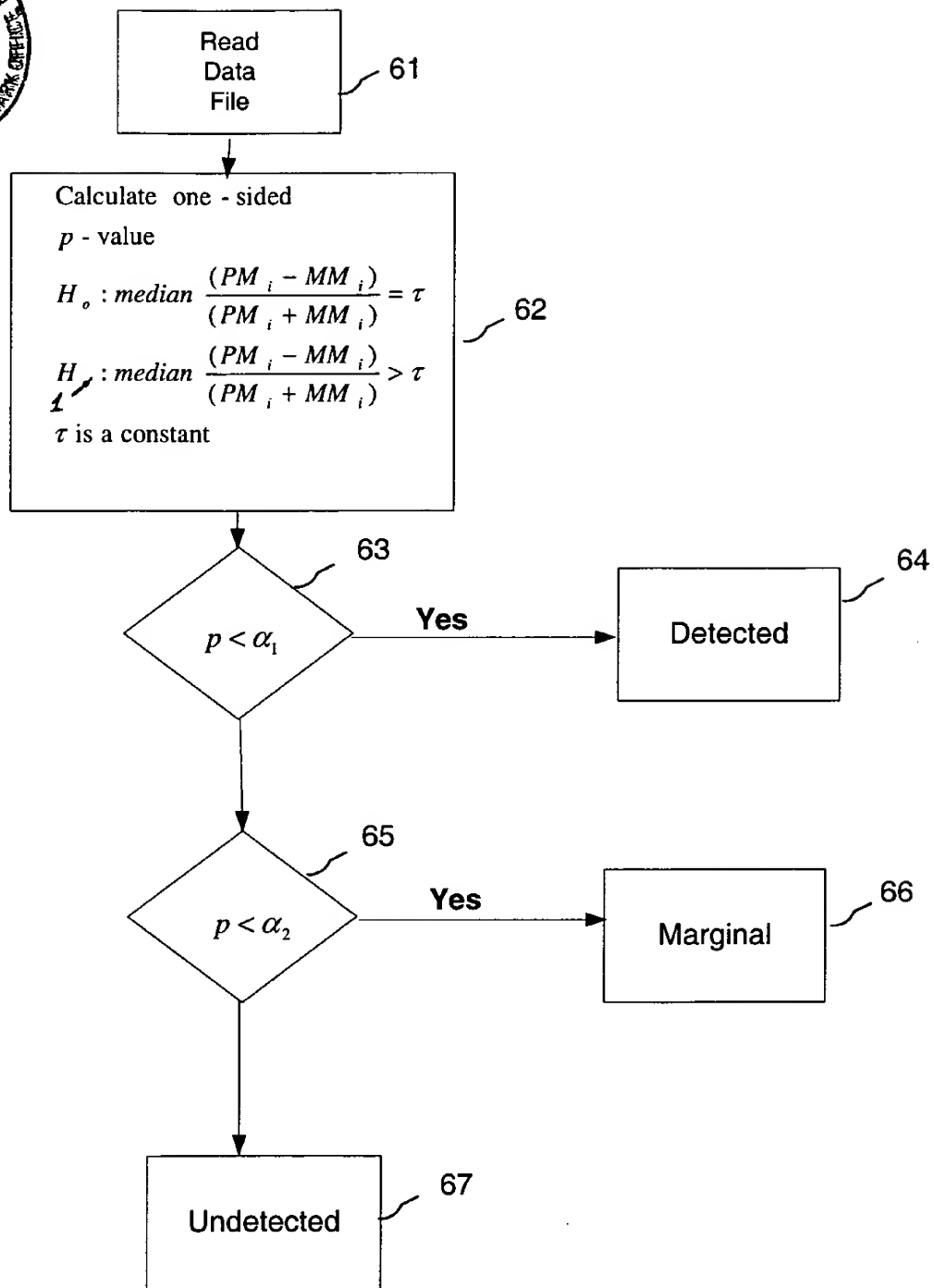
indicating whether said transcript is present based upon said  $p$  value.

103. (new) A method for determining whether a transcript is present in a biological sample comprising:

providing a plurality of perfect match intensity values ( $PM_i$ ) and mismatch intensity values ( $MM_i$ ) for at least 5000 transcripts, wherein the  $PM_i$  for each of said 5000 transcripts is paired with one of the  $MM_i$ ;

calculating a  $p$ -value using one-sided Wilcoxon's signed rank test, wherein the  $p$ -value is for a null hypothesis that  $\theta$ =a threshold value and an alternative hypothesis that said  $\theta$ > said threshold value, wherein said  $\theta$  is a test statistic for intensity difference between said perfect match intensity values and mismatch intensity values; and

indicating whether said transcript is present based upon said  $p$ -value.



Figur 6